

## WHAT IS CLAIMED IS:

1           1. A method for assessment of the intra-amniotic environment,  
2     comprising (A) obtaining a vaginal sample from a subject, and  
3     (B) subjecting the sample to analysis, to determine presence or absence in  
4     the sample of a plurality of biomarkers that is indicative of status of the  
5     intra-amniotic environment, such that results from the assessment of the  
6     vaginal sample informs a diagnostic or prognostic determination in relation  
7     to the subject.

8           2. The method as claimed in claim 1, wherein (A) and (B) are  
9     repeated at least at a second time.

10          3. The method as claimed in claim 1, wherein the biomarkers are  
11     indicative of rupture of the fetal membrane.

12          4. The method as claimed in claim 1, wherein the biomarkers are  
13     indicative of intra-amniotic infection.

14          5. The method as claimed in claim 1, wherein the biomarkers are  
15     indicative of intra-amniotic inflammation.

16          6. The method as claimed in claim 1, wherein the biomarkers are  
17     indicative of fetal lung maturation.

18          7. The method as claimed in claim 1, wherein the biomarkers are  
19     selected from the group consisting of alpha-fetoprotein, fetal fibronectin,  
20     insulin-like growth factor binding protein-1, prolactin and human placental  
21     lactogen, and fragments thereof.

22          8. The method as claimed in claim 1, wherein the biomarkers are  
23     selected from the group consisting of beta-2-microglobulin and cystatin-C,

24 and fragments thereof.

25 9. The method as claimed in claim 1, wherein the plurality of  
26 biomarkers is subjected to pattern recognition analysis.

27 10. The method as claimed in claim 1, wherein the method is an  
28 ELISA.

29 11. The method as claimed in claim 1, wherein the method  
30 comprises mass spectrometric analysis effected via SELDI.

31 12. The method as claimed in claim 11, wherein the method  
32 comprises applying the vaginal sample to a biochip comprising at least  
33 one absorbent selected from the group consisting of a hydrophobic  
34 adsorbent and a cation exchange absorbent.

35 13. The method as claimed in claim 11, the mass spectrometric  
36 analysis comprises subjecting mass-spectrometry peak data obtained for  
37 the vaginal sample to software analysis comprised of an algorithm for  
38 analyzing data extracted from a spectrum.

39 14. The method as claimed in claim 13, wherein the algorithm  
40 implements a pattern-recognition analysis that is keyed to data relating to  
41 at least one of the biomarkers.

42 15. The method as claimed in claim 1, wherein a first vaginal  
43 sample is collected early during a pregnancy and contributes to a baseline  
44 against which subsequent vaginal samples are compared.

45 16. The method as claimed in claim 15, wherein the  
46 determination includes a recommendation for treatment.

47 17. The method as claimed in claim 16, further comprising

48 monitoring the treatment by assaying at least one vaginal sample during  
49 treatment, to determine the presence or absence in the vaginal sample of  
50 biomarkers that are indicative of status of the intra-amniotic environment.

51 18. The method as claimed in claim 16, wherein the  
52 determination includes a recommendation of treatment that comprises  
53 antibiotic treatment, tocolytic treatment, anti-inflammatory treatment, or  
54 antioxidant treatment.

55 19. The method as claimed in claim 16, wherein the  
56 determination includes a recommendation of treatment that comprises  
57 inducing labor.

58 20. The method as claimed in claim 16, wherein the  
59 determination includes a recommendation of treatment that comprises a  
60 cesarean section.

61 21. A method for assessment of the intra-amniotic environment,  
62 comprising (A) obtaining a vaginal sample from a subject, (B) subjecting  
63 the sample to analysis, to determine the presence or absence in the  
64 sample of one or more oxidized or carbonylated peptides that are  
65 indicative of status of the intra-amniotic environment, such that results  
66 from the assessment of the vaginal sample informs a diagnostic or  
67 prognostic determination in relation to the subject.

68 22. The method as claimed in claim 21, wherein the vaginal  
69 sample is treated with dinitrophenol which is incorporated into the  
70 oxidized or carbonylated peptide.

71 23. The method as claimed in claim 21, wherein the method is  
72 an ELISA.

73           24.    The method as claimed in claim 21, wherein the method  
74   comprises mass spectrometric analysis effected via SELDI.

75           25.    The method as claimed in claim 21, wherein the method  
76   comprises applying the vaginal sample to a biochip comprising at least  
77   one absorbent selected from the group consisting of a hydrophobic  
78   adsorbent and a cation exchange absorbent.

79           26.    The method as claimed in claim 24, wherein the mass  
80   spectrometric analysis comprises subjecting mass-spectrometry peak data  
81   obtained for the vaginal sample to software analysis comprised of an  
82   algorithm for analyzing data extracted from a spectrum.

83           27.    The method as claimed in claim 26, wherein the algorithm  
84   implements a pattern-recognition analysis that is keyed to data relating to  
85   a plurality of oxidized or carbonylated peptides.

86           28.    The method as claimed in claim 21, wherein a plurality of  
87   oxidized or carbonylated peptides is subjected to pattern recognition  
88   analysis.

89           29.    The method as claimed in claim 21, wherein a first vaginal  
90   sample is collected early during a pregnancy and contributes to a baseline  
91   against which subsequent vaginal samples are compared.

92           30.    The method as claimed in claim 21, wherein the  
93   determination includes a recommendation for treatment.

94           31.    The method as claimed in claim 30, further comprising  
95   monitoring the treatment by assaying at least one vaginal sample during  
96   treatment, to determine the presence or absence in the vaginal sample of  
97   the one or more oxidized or carbonylated peptides.

98        32.    The method as claimed in claim 30, wherein the  
99    determination includes a recommendation of treatment that comprises  
100    antibiotic treatment, tocolytic treatment, anti-inflammatory treatment, or  
101    antioxidant treatment.

102       33.    The method as claimed in claim 30, wherein the  
103    determination includes a recommendation of treatment that comprises  
104    inducing labor.

105       34.    The method as claimed in claim 30, wherein the  
106    determination includes a recommendation of treatment that comprises a  
107    cesarean section.

108       35.    The method as claimed in claim 22, wherein the treated  
109    vaginal sample is applied to a biochip comprising an anti-dinitrophenol  
110    antibody and subjected to mass spectrometric analysis that is keyed to a  
111    shift in molecular weight, relative to a sample not treated with  
112    dinitrophenol, that corresponds to the incorporated dinitrophenol group.

113       36.    The method as claimed in claim 22, wherein the treated  
114    vaginal sample is applied to a biochip comprising an anti-dinitrophenol  
115    antibody and subjected to mass spectrometric analysis is keyed to a shift  
116    or approximately 16 Da, relative to a sample not treated with  
117    dinitrophenol, that corresponds to the molecular mass of oxygen.

118       37.    The method as claimed in claim 21, wherein total carbonyl  
119    content of the oxidized or carbonylated peptides is measured by  
120    derivatizing the peptides with dintrophenylhydrazine.

121       38.    A method for qualifying status of the intra-amniotic  
122    environment in a subject over time, comprising (i) providing spectra  
123    generated by mass spectrometric analysis of at least two vaginal samples

124 taken from the subject, and (ii) extracting data from the spectra and  
125 subjecting the data to pattern-recognition analysis that is keyed to at least  
126 two peaks in the spectra.

127 39. A kit for detecting, from a sample of vaginal fluid, the  
128 presence of at least two biomarkers indicative of status of the intra-  
129 amniotic environment, comprising (a) a substrate adapted for inserting  
130 into a mass spectrophotometer for analysis, and (b) instructions for  
131 applying a sample of vaginal fluid to the substrate and subjecting the  
132 substrate to mass spectrometric analysis.

133 40. The kit as claimed in claim 39, wherein the substrate is a  
134 biochip.

135 41. The kit as claimed in claim 40, wherein the biochip  
136 comprises at least one absorbent selected from a hydrophobic adsorbent  
137 and a cation exchange adsorbent.

138 42. The kit as claimed in claim 40, wherein the biochip  
139 comprises an anti-dinitrophenol absorbent.

140 43. The kit as claimed in claim 39, additionally comprising, in a  
141 separate container, a quantity of the biomarker in pure form to be used as  
142 a standard.

143 44. The kit as claimed in claim 43, a washing solution for  
144 removing unbound material from the substrate.

145 45. A kit for detecting, from a sample of vaginal fluid, the  
146 presence of at least one oxidized or carbonylated peptide indicative of  
147 status of the intra-amniotic environment, comprising (a) a substrate that  
148 binds the peptide, and (b) instructions for applying a sample of vaginal

149 fluid to the substrate and subjecting the substrate to analysis.

150 46. A kit as claimed in claim 45, comprising an ELISA substrate.

151 47. The kit as claimed in claim 45, comprising a substrate  
152 adapted for insertion into a mass spectrophotometer for analysis.

153 48. The kit as claimed in claim 45, additionally comprising, in  
154 separate container, a quantity of the oxidized or carbonylated peptide in  
155 pure form to be used as a standard.

156 49. The kit as claimed in claim 48, a washing solution for  
157 removing unbound material from the substrate.

158 50. A method for identifying biomarkers that are present in  
159 vaginal fluid and are indicative of status of the intra-amniotic  
160 environment, comprising:

161 (a) profiling a sample of vaginal fluid by mass  
162 spectrophotometric analysis,

163 (b) profiling a sample of amniotic fluid by mass  
164 spectrophotometric analysis, and

165 (c) comparing the profiles obtained in (a) and (b) to identify  
166 biomarkers in vaginal fluid that also are found in amniotic fluid.

167 51. The method as claimed in claim 50, additionally comprising  
168 correlating the presence or absence of the biomarkers in the vaginal fluid  
169 that are also found in the amniotic fluid to a clinical status.

170 52. The method as claimed in claim 51, wherein the clinical  
171 status is rupture of the fetal membrane.

172 53. The method as claimed in claim 51, wherein the clinical  
173 status is intra-amniotic infection.

174        54.    The method as claimed in claim 51, wherein the clinical.  
175    status is intra-amniotic inflammation.

176        55.    A method for identifying biomarkers that are present in  
177    vaginal fluid and are indicative of status of the intra-amniotic  
178    environment, comprising:

179                (a) profiling a first sample of vaginal fluid from a subject  
180    having a normal pregnancy by mass spectrophotometric analysis,

181                (b) profiling a second sample of vaginal fluid from a subject  
182    having a pregnancy characterized by an abnormal clinical status by mass  
183    spectrophotometric analysis, and

184                (c) correlating the presence or absence of the biomarkers in  
185    the vaginal fluid to clinical status of the pregnancy.